Chapter I

General Background

Introduction

Theophylline , a xanthine derivative, is widely used not only as an effective bronchodilator in the treatment of bronchial asthma and other respiratory diseases but also as a prophylactic drug for controlling the symptoms of chronic asthma (Winter, 1982 ; Sa, Bandyopadhyay, and Gupta, 1990 ; Hendeles, Massanari, and Weinberger,1991). Its usual therapeutic plasma concentration is in the range of 10 to 20 mcg/mL (Jenne et al., 1972 ; Mitenko and Ogilvie, 1973 ; Weinberger and Riegelman, 1974). Due to this narrow therapeutic range, careful control of its release from dosage forms has to be ensured to minimize or eliminate any toxic effects that might occur. In addition, the relatively short half-life of theophylline makes the inconvenience for patient to repeatedly take oral conventional dosage form, which may be as frequent as every six hours (Georgarakis et al., 1990). These two significant problems have made theophylline an ideal candidate for administration as sustained or controlled-release preparations. The benefits of such dosage forms are (Buckton, Ganderton, and Shah, 1988) :

(1) to increase patient compliance : a once or twice daily dosage being preferable to 3 or 4 times a day.

(2) to provide a better control of therapeutic drug level : this results in fewer side effects, giving greater likelihood of compliance, and improved disease management.

Sustained-release dosage forms could be prepared as matrix system in which active drug, retardant polymer, and other additives are all uniformly dispersed. A number of methods and techniques have been used in the preparation of oral sustained-release formulations, e.g. direct compression (Ford et al., 1987 ; Hogan, 1989), conventional wet granulation (Klinger et al., 1990 ; Li and Tu, 1991), spray drying (Kawashima and Takenaka, 1974 ; Takeuchi, Handa,and Kawahima, 1989), and polymer coating of drug granule by fluidization (Friedman and Danbrow, 1978 ; Li et al., 1989). The method of manufacture may be complicated depending upon the desired formulation. The example of such product is Theo-Dur®(commercial theophylline sustained-release tablet) which is a combination of coated beads embedded in a slowly disintegrating matrix (Shangraw, 1988). Conventional wet granulation may be regarded to be a simple and convenient method. In spite of this, its disadvantages should be taken into account. By comparison with spray drying, conventional wet granulation may produce less uniform polymer distribution in the matrix especially when the polymer is highly viscous

and thus incorporated with low quantity. This result certainly has a direct effect on the reproducibility of drug release.

Recently, the research team in this laboratory has developed theophylline sustained-release matrices using two different techniques, i.e. spray drying (Vipaluk, 1993) and fluidized-bed coating of drug granules (Sudarat, 1994), both employing ethylcellulose polymer as retarding agent. In vitro testing of those products showed the promising results of the constant and reproducible drug release which were comparable to the market product, Theo-Dur®. In this study, theophylline sustained-release tablets prepared by conventional wet granulation would be developed to obtain the satisfactory release characteristics and then the selected formulation together with others prepared by spray drying (Vipalak,1993) and fluidization coating (Sudarat,1994) would be evaluated for their relative bioequivalences with two commercial products, Theo-Dur® and Nuelin®, in rabbits.

Animal models for oral drug absorption are used for several purposed (Dressman and Yamada, 1991). One of these is to test new formulations for their ability to deliver the drug in a timely and effective manner. Results in animal can be used to aid in selection of the most appropriate oral formulation and as a guide in the design of controlled-release dosage forms. The rabbits are used in this study because they are readily available, relatively inexpensive and easy to house and handle although they have some physiological differences from humans (Maeda

et al.,1977 quoting Chiou et al., 1969). In fact, the purpose of this study is to comparatively evaluate the experimental tablets with reference to Theo-Dur® which its in vivo characteristics has been thoroughly studied in human but is not to extrapolate the animal results to be a generalization of product's efficacy in human.

Objectives of the Study

1. To evaluate theophylline sustained-release tablets prepared by different techniques in rabbits.

2. To develop the formulation of theophylline sustained-release tablets which provide constant drug release over twelve hours by conventional wet granulation method and also study the effect of formulation variables, including the quantity of polymer and channeling agent, and the effect of pH of dissolution medium on drug release.

3. To study the bioavailability and pharmacokinetics of theophylline sustained-release tablets prepared by conventional wet granulation, spray drying, and fluidized-bed coating in rabbits.

4. To assess the bioequivalences among the experimental tablets themselves and with Theo-Dur® and Nuelin® by statistical analysis of the relevant pharmacokinetic parameters, C_{max}, t_{max}, and AUC_0^∞.

5. To investigate the correlation between the dissolution rate constant and the bioavailability parameters, C_{max}, t_{max}, and AUC_0^∞.

Literature Reviews

1. Theophylline

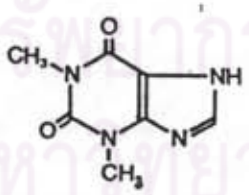
Theophylline is designated by Chemical Abstracts as 1,3-dimethylxanthine. It is also known as theocin. It is similar in structure to the common dietary xanthines, i.e. caffeine and theobromine. Although theophylline presents in coffee, tea, and cocoa as well as other natural sources, it is made available commercially by total synthesis.

1.1 Physicochemical Properties(Cohen, 1975;Swinyard, 1990)

Chemical name : 3,7-Dihydro-1,3-dimethyl-1H-purine-2,6-dione

Empirical formula : $C_7H_8N_4O_2$

Structural formula :



Molecular weight : Theophylline monohydrate 198.18

Theophylline anhydrous 180.17

Description	:	White, odorless, crystalline powder with a bitter taste. Saturated aqueous solution is neutral or slightly acid to litmus.
Solubility	:	8.3 mg/mL in water, 12.5 mg/mL in ethanol, 11.6 mg/mL in chloroform, sparingly soluble in ether, and freely soluble in solutions of alkali hydroxides and ammonia.
Melting range	:	271-274 ° C.
pK_a	:	Theophylline is a weakly acidic compound with relatively high pK_a of 8.6.
Stability	:	Stable in air. Solutions are generally quite stable over the entire pH range. Strongly alkaline solutions (pH > 12) show decomposition after several weeks. Precipitation from aqueous solution will occur if the pH drops below 9 unless presents in concentrations less than the water solubility.

1.2 Therapeutic and Toxic Effects

For decades theophylline has been recognized as a potent bronchodilator for the relief of acute asthmatic symptoms. It is also used as a respiratory stimulant for Cheyne-Stokes respirations and as an adjunct in the treatment of acute pulmonary edema. More recently it has been used to prevent episodes of idiopathic apnea or bradycardia in premature newborns. Its most important current use, however, is as a major "prophylactic" agent for controlling the symptoms of chronic asthma. Application of new knowledge related to the pharmacodynamic and pharmacokinetic characteristics of theophylline and the availability of rapid specific serum assays have improved both the efficacy and safety of this drug. In addition, reliably-absorbed, slow-release formulations have been developed that provide a highly effective and convenient means of maintaining around-the-clock stabilization of the hyper-reactive airways that characterize chronic asthma (Hendeles et al., 1991).

Several studies have proved that the clinical response to theophylline is greatly related with its plasma concentration. the bronchodilator effect in hospitalized patients with reversible airway obstruction is shown to be proportional to the logarithm of the plasma concentrations over the range of 5-20 mcg/mL (Mitenko and Ogilvie, 1973 ; Levy and Koysooko, 1975). Pulmonary function also improves or decreases in the manner that is roughly the same trend with the rise and fall in the plasma concentration (Jenne et al., 1972 ; Simons, Luciuk, and

Simon, 1982). Inhibition of exercise-induced bronchospasm, a significant manifestation of asthma, is found to relate to plasma concentration with clinically important effects most apparent above 10 mcg/mL and even greater effects above 15 mcg/mL (Pollack et al., 1977). Prevention of the signs and symptoms of chronic asthma has also been demonstrated when plasma concentrations are maintained within the 10-20 mcg/mL range on the continuous around-the-clock basis (Weinberger and Bronsky, 1974).

The adverse side effects as well as toxicity of theophylline are also appeared to have a relationship to plasma concentration. Nausea and vomiting are the most common side effects. Although these effects can occur at concentrations as low as 13 to 15 mcg/mL, they are observed more frequently at plasma concentrations exceeding 20 mcg/mL (Jenne et al., 1972 ; Jacobs, Senior, and Kessler, 1976). Adverse effects associated with serum concentrations above 20 mcg/mL includes nausea, vomiting, headache, diarrhea, irritability, and insomnia (Jacobs et al., 1976). At higher plasma levels (i.e., > 35 mcg/mL), hyperglycemia, hypokalemia, hypotension, cardiac arrhythmias, hyperthermia, seizures, brain damage, and death may occur (Zwillich et al., 1975 ; Hendeles et al., 1977).

1.3 Commercial Theophylline Sustained-release Products

There has been a wide variety of theophylline sustained-release products commercially available today since the first modern one, Aerolate®, was marketed by Fleming and Co. in 1972-1973 (Shangraw,

1988). Most of these products have the different composition and release mechanism. Here, however, is a discussion of some commercial products available in Thailand.

1.3.1 Nuelin®SR, 250 mg (Riker Laboratories) : It consists of waxy non-disintegrating bed, the surface of which is coated with cellulose acetate. The mechanism of drug release from tablet solely undergoes surface erosion (Buckton, Ganderton, and Shah, 1988).

1.3.2 Quibron® (Mead Johnson Lab) : This may be regarded to be one of the most unique sustained-release products of theophylline as it contains almost no excipients (95 % drug). The patent describes a granulation of anhydrous theophylline with 5 % hydroxypropyl methylcellulose which is finally lubricated with 0.5 % magnesium stearate and compressed (David, Brooke, and Gallian, 1984). Upon exposure to body fluids, the hydrophilic polymer begins to hydrate and swell into a gelatinous mass. The drug will slowly dissolve and then diffuse through the gel barrier without tablet disintegration. In addition, the plate-like geometry of tablet also results in a gradual change of surface area with time providing a more uniform dissolution rate. However, it was shown in the recent study of Phuriwat (1991) that the complete in vitro drug release from Quibron® was achieved at only about 6 hours.

1.3.3 Theo-Dur® (Key Pharmaceuticals) : This is certainly the most successful of all the sustained-release theophylline products. Its relatively uniform release pattern over 12 to 18 hours together with commercial popularity has made Theo-Dur® the standard against which all other sustained-release theophylline products are compared. On the other hand, it is one of the most complicated products in terms of both formulation and method of manufacture. Theophylline is coated onto sugar beads which are then enclosed in various coatings of lipid material (glyceryl monostearate, cetyl alcohol, beeswax) and / or an acid polymer cellulose acetate phthalate. The beads are then blended with the base which contains a fraction of the dose as well as excipient and finally compressed into a slowly disintegrating matrix. Drug release depends on the type and thickness of the coatings, nature of the tablet matrix, and also the geometry and hardness of the tablets. In spite of its complexity, Theo-Dur® has established a record of constant drug delivery, which is not substantially affected by food and pH (Gonzalez and Golub, 1983 ; Shangraw, 1988).

1.3.4 Theo-24® (Searle Pharmaceuticals) : It is the first product introduced into the pharmaceutical market for 24-hour dosage interval. Since its introduction there has been considerable controversy as to the appropriateness of such a dosing schedule in the regulatory agencies, the medical community, and the courts. Although the promotional literature describing the sustained-release pellets (Probeads®) is quite elaborate, the company does not provide any information on the

composition of the product or to explain how the beads are designed to "prolong transit time". It does appear to be a diffusion controlled coated bead type product which utilizes either shellac or cellulose acetate phthalate and which is pH dependent. Subsequent studies have shown that it releases theophylline at a more rapid rate when given with food than taken on an empty stomach (Shangraw, 1988).

2. Hydroxypropyl Methylcellulose (HPMC)

HPMC is widely used in the controlled-release formulations as the basis for hydrophilic matrices. In this study, a high viscosity grade of HPMC, Methocel E4M, is used as retarding agent in the production of theophylline sustained-release tablets.

2.1 Physicochemical Properties

HPMC is a propylene glycol ether of methyl cellulose. It is an odorless, tasteless, white or creamy-white fibrous or granular powder. It is soluble in cold water forming a viscous colloidal solution ; insoluble in alcohol, ether and chloroform ; but soluble in mixtures of methyl alcohol and methylene chloride. HPMC is very stable in dry conditions as its solutions are stable at pH 3.0-11.0. The incompatibility may occur in extreme pH conditions or with oxidizing materials. Human and animal feeding studies have proved HPMC to be safe (The Pharmaceutical Society of Great Britain, 1986).

As described in the USP XXII (1990), HPMC is classified into four types according to the percentage of methoxy ($-OCH_3$) and hydroxypropoxy ($-OCH_2CHOHCH_3$) groups after being dried at 105 °C for two hours. The standard limits on content of substituents for each type are set forth in Table 1.

Table 1 Classification of Hydroxypropyl Methylcellulose

Substitution Type*	Methoxy (percent)		Hydroxypropoxy (percent)	
	Min.	Max.	Min.	Max.
1828	16.5	20.0	23.0	32.0
2208	19.0	24.0	4.0	12.0
2906	27.0	30.0	4.0	7.5
2910**	28.0	30.0	7.0	12.0

* Of the four digits in each number, the first two define the average percent content of methoxy groups and the last two define that of hydroxypropoxy groups.

** the commercial example for this type of HPMC is Methocel E4M which has the viscosity of about 4000 cP.

2.2 Applications in Matrix Type Sustained-release Formulation

Hogan (1989) has prepared the matrix type sustained-release tablets of some drugs using different grades of Methocel (commercial product of HPMC) by direct compression and investigated the relationship between release rate and quantity of polymer. It was concluded that an increase of the fraction of Methocel in the tablet could retard the rate of drug dissolution. The principle, as described by the researcher, for controlling drug release in the matrix tablet is that, on exposure to aqueous fluids, the tablet surface becomes wet and the polymers starts to partially hydrate to form a gel layer. An initial burst of soluble drug from the external layer may be released. There follows an expansion of the gel layer and soluble drug diffuses through the gel barrier. Concomitantly the outer layer becomes fully hydrated and dissolves, a process generally referred to as erosion. Water continues to penetrate towards the tablet core until it has completely dissolved.

In 1990, Sanghavi, Kamath, and Amin have prepared the sustained-release tablets of theophylline using either hydrophilic polymers (methylcellulose, Methocel K4M and K15M) or hydrophobic polymers (ethylcellulose, cellulose acetate) by means of wet granulation. The result of increasing Methocel quantity was the same as discovered by Hogan (1989). Furthermore, it was found that the matrix tablets with only

Methocel K4M exhibited non-Fickian diffusion whereas those prepared using Methocel K15M followed zero-order kinetics.

Alderman (1984) also described the prolonged release from HPMC matrices and concluded that a gelatinous layer, formed when the polymer hydrated on contact with water, controlled the release of drugs by two mechanisms. Water soluble drugs were released by diffusion out of the gelatinous layer and by erosion of the gel, whereas poorly soluble drugs were released only by erosion.

The influence of various formulation factors on drug release from HPMC matrix tablets was fully examined by Ford et al (1985 a, b, c). They described that the major factor controlling drug release was the drug : HPMC ratio and for each of the drugs a straight line relationship existed between the release rate and the reciprocal of the weight of HPMC in the matrices.

Other applications of HPMC in pharmaceutical formulation are as follows (The Pharmaceutical Society of Great Britain, 1986) :

1. Film former in tablet film coating (perhaps the most commonly used film-forming agent).

2. Lower viscosity grades are used in aqueous film coating and higher viscosity grades are used in solvent film coating. The concentration varies from 2 to 10 % depending upon the viscosity grade of the polymer.

3. Binder in tablet granulation at 2 to 5 % .

4. Thickening agent added to vehicles for eye drops and artificial tear solutions at 0.45 - 1.0 % .

5. Protective colloid which prevents droplets and particles from coalescing or agglomerating, thus inhibiting the formulation of sediments.

6. Emulsifier, suspending agent and stabilizer in gels and ointments.

7. Adhesive in plastic bandages.

3. Channeling agent

The term " channeling agent " has initially been used by Sanghavi et al (1990). It is a group of highly water-soluble substances, such as PEG 4000, polyvinyl alcohol (PVA), polyvinylpyrrolidone (PVP), which is able to increase the dissolution rate of slightly soluble drug from sustained-release matrices when mixing at an adequate quantity. This may be explained by the rapid dissolution of the channeling agent in the aqueous medium which consequently forms many pores in the tablet.

This is equal to an increase of surface area available for the drug to dissolve. In addition, the soluble drug could be directly released by these pores as well as the diffusion through the hydrated polymer.

These materials might be called " porosigen " according to the research of Cararelli (Baker, 1987) in which ammonium sulfate and lactose were used to modify drug release.

4. In Vitro-In Vivo Correlations

Once the utility of controlled-release dosage form was established, numerous methods were developed to achieved extended-release with solid oral dosage forms (Lazarus and Cooper, 1961; Robinson, 1978). These methods are based upon the principles of diffusion, erosion, chemical reaction, osmotic pressure, or some combination theoref and utilize both natural products (waxes, gelatin, modified protein) and synthetic materials (cellulose esters, polymers, ion exchange resins). However, regardless of the method used to achieve the desired result, the process employed to develop a product with acceptable in vitro characteristics usually consists of the following four steps (Leeson et al., 1985) :

(1) Developing one or more formulations which demonstrate a slow release pattern in vitro.

(2) Studying the system(s) from step 1 in vivo and evaluating the resulting plasma level curves.

(3) Accepting a dosage form if it produced what is defined as an acceptable plasma level curve.

(4) Returning to step 1 if no dosage form is considered to be acceptable.

To avoid the costly in vivo screening approach described above, several efforts have been made to establish some form of correlation between in vitro and in vivoparameters which is then utilized to predict the in vivo performance of a formulation (Wiegand and Tayler, 1960; Robinson and Eriksen, 1966; Smolen, 1971; Vaughan and leach, 1976; Aarons and Rowland, 1977). From these predictions or simulation, the dosage form with the desired characteristics would be selected for further in vivo evaluation. The advantages of establishing such a relationship are to be measured in terms of cost, time, and safety. For an in vitro test to be useful in this context, it must predict in vivo behavior to such an extent that an in vivo bioavailability test becomes redundant.

In vitro specification such as physical and chemical properties, stability, water content, disintegration, solubility, and the rate and extent of dissolution are routinely used as quality and process controls in dosage form manufacturing. These characteristics are well established, and it is

tempting to consider that one or more of them may be useful to predict behavior of the dosage form in the GI tract and its overall absorption characteristics.

Recently, dissolution rate has been used as a manufacturing process standard and is generally considered to be the in vitro parameter most likely to correlate with in vivo bioavailability which is described in terms of the rate and the extent of drug absorption. Rate of absorption is reflected in peak drug concentrations in plasma (C_{max}) and the times at which they occur (t_{max}). Extent of absorption is reflected in C_{max} and also the area under the plasma drug curve (AUC). Therefore, one has the quandary as to which term(s) to select in seeking an in vitro-in vivo relationship. Accurate correlation, if it is achievable, will depend on accurate selection of the in vitro parameter that has the greatest intrinsic effect on drug absorption characteristics. Usually, the in vivo parameters C_{max} and AUC are compared with in vitro dissolution rate. However, both of these in vivo parameters are functions of both the rate and extent of absorption ; consequently, they may be influenced as much by solubility as by dissolution rate. Thus, depending on the site(s) at which drug is absorbed from the GI tract, solubility and dissolution rate may have varying effects on the drug-absorption profile.

In the case of oral controlled-release dosage forms, the complete plasma level-time curve is of greater importance since the shape of these curves is critical in defining product performance (Leeson et al.,

1985). Therefore, relating the in vitro dissolution behavior of a sustained-release dosage form to the complete plama level curve represents the type of correlations that would be of interest to both the formulator and clinical pharmacologist.

However, the characteristics of such dosage forms is found to present additional complications. They are designed to release drug slowly in the GI tract by employing a variety of mechanisms such as coating, wax matrices, osmotic pump devices, and others. Because each of these dosage forms is unique, it would not be expected that a single dissolution test could accurately predict drug absorption from these dosage forms in the complex environment of the GI tract. One suggestion for a controlled-release dosage form is that a variety of formulations, with varying dissolution characteristics, be prepared for each product. Bioavailability studies should then be conducted to establish in vitro-in vivo relationship (Welling, 1991).

A few studies are performed to investigate the in vitro-in vivo correlation of theophylline sustained-release formulations. In 1985, El-Yazaki and Sawchuk determined parameters of theophylline from six commercial products (three uncoated and three sustained-release formulations) and also investigate correlations between the data obtained from the dissolution (e.g. % of drug dissolved in 30, 60 minutes; and dissolution rate constant) and absolute bioavailability studies in the rabbit. The good correlations were obtained ($r^2 > 0.9$) and can be reliably

employed to predict bioavailability parameters from in vitro dissolution variables.

Another research conducted by Hussein and Friedman (1990) is a development of five experimental theophylline sustained-release formulations in a matrix tablet form with a protein carrier. In comparison with Theo-Dur® and Theotrim®, the in vitro drug release from these tablets were studied and the selected formulations were then evaluated for their pharmacokinetics in dogs and human. For each of the formulation tested, the regression analysis results of the percentage of theophylline absorbed against the mean percentage released in vitro, at the corresponding times, indicated a high correlation. These data imply that the drug release profiles in the GI tract of dogs and human under fasting conditions and those in vitro are similar and that theophylline release is the rate-limiting step in its absorption.